HiPrep IMAC FF Columns

Product Information

Cat#No#	Hi-063P

Product Overview

HiTrap IMAC FF is prepacked with IMAC Sepharose 6 Fast Flow. The resin is charged with the metalion of your choice for Immobilized Metal ion Affinity Chromatography (IMAC) and subsequent purification of polyhistidine tagged proteins.

Description

IMAC Sepharose FF is supplied free of metal ions. It is charged by the user with the transition metal ion of choice (e.g. Cu2+, Zn2+, Ni2+, or Co2+); these metal ions will bind to the covalently immobilized chelating ligand on the Sepharose.

Characteristic

For optimizing purification of histidine-tagged proteins when Ni2+ is not the best choice of metal ion.
Conveniently charge with your metal of choice.
Prepacked 20 mL HiPrep columns for easy scale-up.
High binding capacity.

Maximum operating pressure

1.5 bar [0.15 MPa] (22 psi)

Sample preparation

Centrifuge at 10 000 × g (or higher) for 10 min and/or filter the sample through 0.45-µm filter. If possible, dilute the sample in binding buffer. The sample should contain imidazole at the same concentration as in the binding buffer.

Metal ion capacity

Approx. 15 µmol Ni2+ /ml medium

Matrix

Highly cross-linked 6% agarose

Average particle size

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CREATIVE BIOMART®
Chromatography

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90 µm
Dynamic binding capacity
Approx. 40 mg (histidine)6- tagged protein/ml medium (Ni2+ - charged). Untagged protein: Approx. 25 mg/ml medium (Cu2+ charged), or approx. 15 mg/ml medium (Zn2+ or Ni2+ charged).
Recommended flow rate
< 300 cm/h
Chemical stability
1 M NaOH, 70% acetic acid. Tested for 12 h. 2% SDS. Tested for 1 h. 30% 2-propanol. Tested for 30 min.
pH working range
2–14
CIP stability
3–12
Storage
4 to 30°C, 20% Ethanol
Binding buffer
20 mM sodium phosphate, 500 mM NaCl, 5 mM imidazole, (1 mM for untagged protein) pH 7.4.
Elution buffer
20 mM sodium phosphate, 500 mM NaCl, 5 mM imidazole, (1 mM for untagged protein) pH 7.4.
Cleaning-in-place
Removal of ionically bound substances: Wash with approximately 20 ml 1.5 M NaCl. Then wash the column with approximately 200 ml distilled water. Removal of precipitated and/or hydrophobically-bound substances and lipoproteins: Wash with 1 M NaOH, contact time usually 1–2 h (longer time may be required to inactivate endotoxins); then wash with approximately 200 ml binding buffer, followed by 100–200 ml distilled water. Removal of hydrophobically-bound proteins, lipoproteins, and lipids: Wash with 100–200 ml 30% isopropanol

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for at least 15–20 min; then wash with approximately 200 ml distilled water. Alternatively, wash with 40 ml detergent in a basic or acidic solution. Use, for example, 0.1–0.5% nonionic detergent in 0.1 M acetic acid, contact time 1–2 h. After treatment, always remove residual detergent by washing with at least 100 ml 70% ethanol. Then wash with approximately 200 ml distilled water.

Pack size
20 mL
Column volume
20 ml
Column i.d.
16 mm
Column hardware pressure limit
0.5 MPa, 5 bar, 73 psi

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